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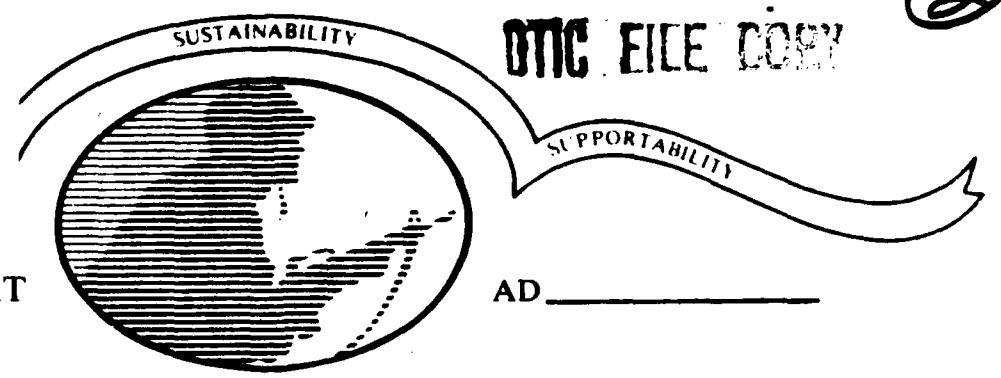

TECHNICAL REPORT
NATICK/TR-87/039

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PRELIMINARY EVALUATION OF THE CONTROL OF MICROBIAL FOULING BY LABORATORY AND PILOT-SCALE AIR-STRIPPING COLUMNS

BY

DARRELL SEEKINS AND MORRIS R. ROGERS

FINAL REPORT

1 MARCH 1985

FOR THE PERIOD
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In order to evaluate biogrowth control in treatment processes, specifically microbial fouling of air-stripping columns, a study was undertaken on pilot and laboratory scales to detect the presence of microbial growth, identify the organisms involved, and to investigate means of preventing fouling.

The pilot-scale column utilized ceramic packing material which was disinfected by super-chlorination prior to use. After three weeks of continuous operation, this column was found to have developed a heavy slime layer caused by ferric hydroxide. The biogrowth on this column was found to be very low.

The laboratory-scale column, which was operated during the daytime only, utilized ceramic packing material that had been previously used in a pilot-scale column and had experienced fouling. The packing material had not been disinfected before use and the column was found to have developed a heavy fouling layer, which was a combination of microbial growth and ferric hydroxide. The predominant microorganisms responsible were; Pseudomonas,

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Bacillus, and Azotobacter.
Treatment of the laboratory-scale column with 3% and 15% hydrogen peroxide reduced total microbial counts but was not successful at disinfecting the column.

It was concluded that to prevent microbial fouling of air-stripping columns, the packing material should be disinfected prior to use and should be shock-chlorinated during use if microbial fouling should start to occur. The column should be run continuously if possible to keep the inside temperature as low as possible to retard the growth of microorganisms. If iron fouling should occur, the column should be treated with dilute HCl to clear the packing material of the hydroxide buildup.

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PREFACE

In December 1983, the U.S. Army Natick Research and Development Center (now known as the U.S. Army Natick Research, Development and Engineering Center (Natick)) undertook a study for the U.S. Army Toxic and Hazardous Materials Agency (USATHAMA) to investigate the buildup of microbial slimes in Air Stripping Columns that are used by the military to remove volatile compounds from contaminated groundwater. This study was performed under USATHAMA Project No. R91. entitled "IR DeCON Technology Program, subtask Biogrowth Control in Treatment Processes", Natick Project No. 1L162704AF25AFBG000.

The Air-Stripping and Carbon Adsorption columns, which were the property of Environmental Science and Engineering (ESE) Inc., Gainesville, FL, were previously used at the Anniston Army Depot (ANAD), Anniston, AL to treat groundwater that had been contaminated with chemical solvents and metal plating wastes. The major groundwater contaminants were; trichloroethylene (TCE), dichloroethylene (DCE), methylene chloride (MeCl), phenol, and chromium.

The results of the study will provide the basis for recommendations on preventative or control measures to be taken in future applications of these water treatment methods. Measures to prevent buildup of microbial slimes will allow for longer column life and reduction in costs.

We wish to thank Micheal D. Cummins and O. Thomas Love, Jr. at Merle Labs, United States Environmental Protection Agency (USEPA) Office of Drinking Water, Cincinnati, OH, and William R. Beckwith, Environmental Science and Engineering, Inc, Gainesville, FL, for their helpful communications and technical advice.

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A PRELIMINARY EVALUATION OF THE CONTROL OF MICROBIAL FOULING BY LABORATORY AND PILOT-SCALE AIR-STRIPPING COLUMNS

INTRODUCTION

In today's industrial society and in various military applications, the use of volatile organic solvents is widespread. The common solvent trichloroethylene (TCE), has been used for years by the military to clean the exterior surfaces of aircraft. Along with the use of solvents comes the problem of environmental pollution and disposal. Numerous groundwater sources have been discovered to be contaminated with TCE or other volatile compounds. Many of these organic solvents are known to be carcinogenic or otherwise hazardous to human health. Others may be potentially dangerous in increased concentrations, or may be found to be hazardous with long-term exposure even at low concentrations.

Since many organic solvents are volatile, the concentrations found in surface waters are generally low, usually in the microgram per liter (µg/L) range.[1] This concentration is very difficult, if not impossible, to detect without sophisticated techniques that were unavailable until recent years.

Various technologies have been developed to deal with the removal of the volatile organic compounds (VOCs) from contaminated water supplies. The most common methods of removal employed today are air-stripping (aeration),[1-7] activated carbon adsorption,[5,6,8,9] and resin adsorption.[10]

The purpose of this investigation was to determine the source of microbial fouling of air-stripping columns and to investigate means of

correcting or preventing slime production to prolong column life.

The model system used for this investigation was one in which an air-stripping column was used to remove trichloroethylene (TCE) from contaminated well water.[7]

MATERIALS

Chemicals: Trichloroethylene (Aldrich chemical, Milwaukee, WI), sodium thiosulfate (J.T. Baker, Phillipsburg, NJ), 30% hydrogen peroxide (Fisher Scientific Co., Fair Lawn, NJ), catalase (Nutritional Biochemicals Corp., Cleveland, OH), sodium hydroxide (J.T. Baker, Phillipsburg, NJ), potassium dichloroisocyanurate (Dorex Inc., Frankfort, IL), potassium iodide starch paper (Arthur H. Thomas Co., Philadelphia, PA), ethanol (Mallinckrodt, Paris, KY), and potassium phosphate monobasic (Fisher Scientific Co., Fair Lawn, NJ).

Media and Reagents: Plate count agar (Difco Laboratories, Detroit, MI), lauryl tryptose broth (Difco Laboratories, Detroit, MI), motility medium (Analytab Products, Plainview, NY), brain heart infusion agar (Difco Laboratories, Detroit, MI), potato dextrose agar (Difco Laboratories, Detroit, MI), mannitol mineral salts agar, starch agar (Difco Laboratories, Detroit, MI), nitrate broth (Difco Laboratories, Detroit, MI), Gram stain reagents (Carr Scarborough Microbiologicals, Stone Mountain, GA), oxidase reagents (Analytab Products, Plainview, NY), nitrate reduction test reagents, Nesslers reagent, Kovacs reagent, triple sugar iron agar (Difco Laboratories, Detroit, MI), and API20E$^{(R)}$ diagnostic test kits (Analytab Products, Plainview, NY).

Laboratory-Scale Air-Stripping Column: One 11-in x 3 3/4-in (27.94-cm x 9.53-cm) glass column, steel spring inside column, 1/4-in (0.64-cm) ceramic "intalox" saddles, 2 clamps, 1 large ringstand, 1/4-in (0.64-cm) glass tubing, 1/2-in (1.27-cm) glass tubing, 1/4-in (0.64-cm) rubber tubing, #4 rubber stopper, 3 3/4-in (9.53-cm) diameter funnel shaped reducer, 3 3/4-in (9.53-cm) diameter stainless steel water distributer, 3 3/4-in (9.53-cm) diameter support screen, air flowmeter, water flowmeter, metering pump (Mec-o-meter, Ecodyne Corp., Miami, FL).

Pilot-Scale Air-Stripping Column: Three 2-ft x 6-in (60.96-cm x 15.24-cm) glass column sections, Teflon^{R} lined gaskets, flanges, flange collars, air inlet tube, support plate, 1/4-in (0.64-cm) ceramic packing, 1-in (2.54-cm) ceramic packing, liquid feed pump, air compressor, water flowmeter (Aquamatic, Rockford, IL), air flowmeter (Aquamatic, Rockford, IL), (1/2-in (1.27-cm) copper) liquid influent piping, (1/2-in (1.27-cm) copper) liquid effluent piping.

Miscellaneous Equipment: Anaerobic incubation jar, thermometer, forceps, vortex mixer (Scientific Industries Inc., Bohemia, NY), turbidity test kit (Hach Co, Ames, IA), hardness test kit (Hach Co., Ames,IA), gas chromatograph equipped with a purge and trap sampler (Hewlett Packard Model 5840A), dissolved solids meter (Lamotte Chemical Products Co., Chestertown, MD), pH meter (Corning Science Products, Medfield, MA), amperometric titrator (Wallace and Tiernan, Bellville, NJ), color test kit (Hach Co., Ames, IA), 120-mL milk dilution bottles, 100 x 25-mm test tubes, 115-mL disposable millipore 0.45μm filters (Millipore corp, Bedford, MA), Nessler tubes (50-mL calibrated), plant grow lights (Sears Roebuck, Chicago, IL), and interval timer (Sears Roebuck, Chicago, IL).

METHOD

To test for fouling, the air-stripping columns were operated for a period of months and periodically tested for microbial growth or slime production. The characterization of the water, and visual inspections of the columns were performed weekly to detect changes as the columns were operated over time. The influent water was spiked with TCE at 100 µg/L to simulate contaminated well water.

Laboratory-Scale Air-Stripping Column:

At the onset of the project, the air-stripping column that was to be used was a 6-ft x 6-in (182.88-cm x 15.24-cm) glass column on loan from Environmental Science and Engineering, Inc. (ESE), Gainesville, FL. When prolonged delays in site location and preparation interfered with the operation of this column, the 11-in x 3-3/4-in (27.94-cm x 9.53-cm) laboratory-scale column shown in Fig. 1 was assembled inside a laboratory fume hood to provide interim data.

The column was supported by two chain clamps attached to a ring stand. The packing material for the column was 1/4-in (0.64-cm) ceramic "Intalox" saddles which were on loan from ESE to be used in the 6-ft x 6-in (182.88-cm x 15.24-cm) column. The saddles were supported in the column by a 3 3/4-in (9.53-cm) diameter screen made of 304 stainless steel with a 0.060-in hole diameter and 0.023-in wire diameter. The screen was located approximately 2-in (5.08-cm) from the bottom of the column and rested on a steel spring which was covered with silicon sealant to prevent rusting. The column was packed with the saddles for 7in above the support screen. On top of the packing rested a 3-3/4-in (9.53-cm) diameter stainless steel plate with 5/8-in (1.59-cm) holes in it to serve as a distributer for the influent water.

On the bottom of the glass column, using a flange clamp, was attached a rounded funnel shaped reducer. The reducer had a 3/4-in (1.91-cm) opening on the bottom into which was inserted a #4 rubber stopper containing two holes. One hole was 1/2in (1.27cm) in diameter and had a 4-in (10.16-cm) long piece of glass tubing inserted into it. The tubing was bent at a 90^{o} angle and served as the effluent port for the column. The other hole had a 1/4-in (0.64-cm) diameter glass tubing inserted into it that served as the air influent port. The top of the air inlet tube was bent at 90^{o} to prevent water from flowing down inside and it extended up to within approximately an inch below the support screen.

The air for this column was fed from the in-house tap line through Tygon$^{(R)}$ tubing via four air flowmeters (Fisher-Porter Co.) that were connected in parallel. Each flowmeter had a capacity of 22 liters/min ($1342.44 in^3/min$).

The influent water for the laboratory scale column was local tap water--the town of Natick uses well water as the public water supply-- and flow was monitored with a "Gilmont model E6677" water flowmeter.

The column was spiked with TCE by pumping a constantly stirred solution containing 0.09 mL/L TCE into the stream of influent water entering the top of the column.

The column was operated for eight hours per day, five days per week using air and water volumes of 66 and 1.5 liters per minute, respectively.

The effluent water hose was raised to a level required to provide a water trap below the air inlet. The trap insured that the air traveled upward through the column rather than out through the effluent water hose. In order to simulate the effect of sunlight, two fluorescent plant growlights were

attached to each side of the column (not shown in Fig 1.). These lights were placed approximately 12in (30.48cm) from the column and were operated for twelve hours per day by means of an interval timer.

Pilot-Scale Air-Stripping Column:

The 6-ft x 6-in (182.88-cm x 15.24-cm) air-stripping column shown in Fig. 2, consisted of three two-foot sections of glass column connected together by using flanges. The column was packed with approximately 5-ft (152.40-cm) of 1/4-in (0.64-cm) ceramic saddles that were supported by a gas-injection-type ceramic plate and a 4-in (10.16-cm) layer of 1-in (2.54-cm) ceramic saddles. At the bottom of the column below the support plate, was a 1-ft (30.48-cm) section of glass column that served as a water trap to prevent air from existing through the water effluent port. A stainless steel liquid distributor was attached at the top of the column.

The water used for this column was the town of Natick well water obtained via the tap. The air used for this column was also supplied via the in-house tap line.

The column was operated continuously for four weeks using a water flow of 1 gallon (3.78 liters) per minute and air flow of 6 SCFM (169.90 liters/min).

The original plans for this column included a TCE spike of the influent water. As a safeguard against backflow of spiked water into the general water supply, a reservoir tank was incorporated into the design to serve as the feed for the column. When the float switch in the reservoir was found to be faulty, the TCE spike was eliminated from the design.

Water Characterization:

The influent waters for the columns were characterized weekly on the basis of the following criteria; turbidity, hardness, dissolved solids, chlorine, pH, color, and temperature.

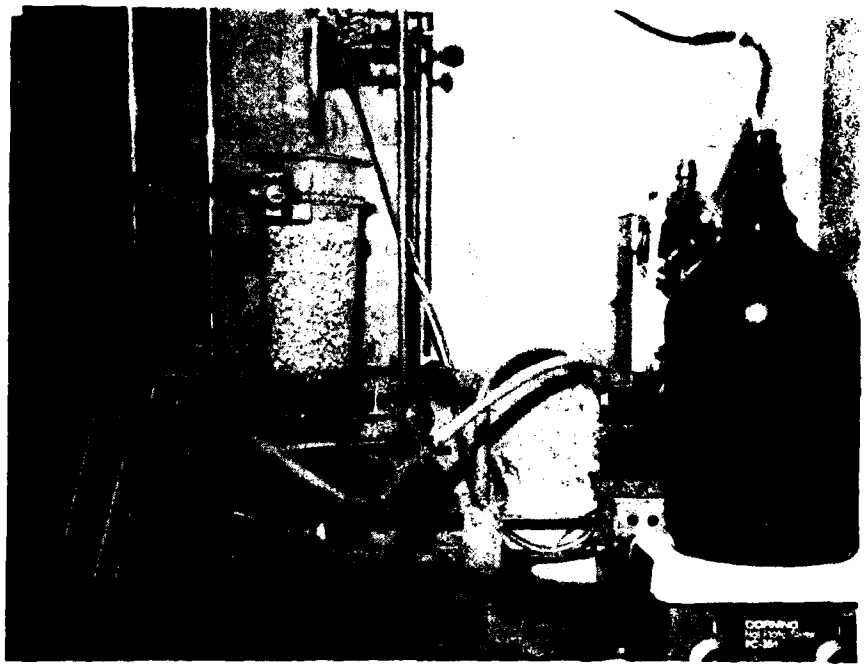

Figure 1. Laboratory-scale air-stripping column

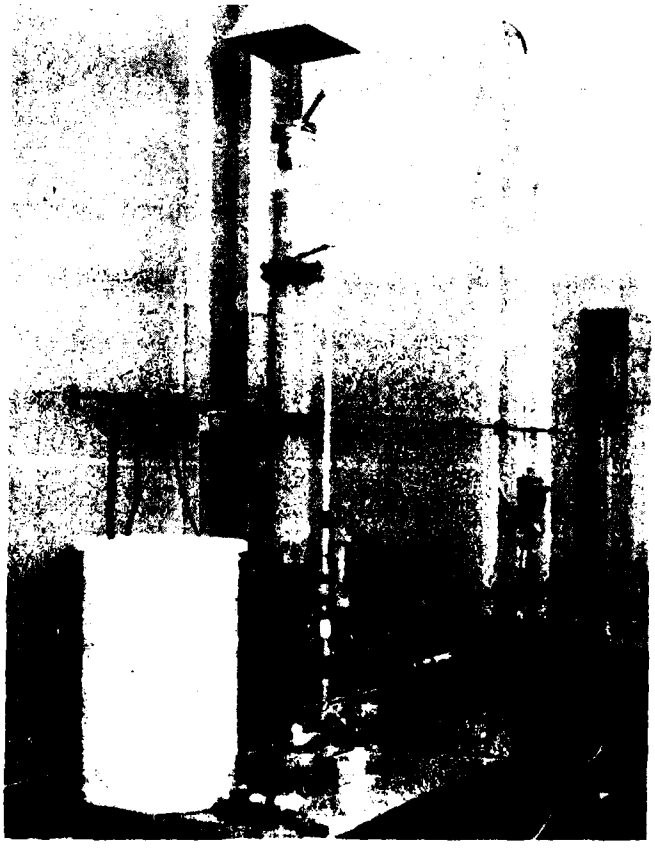

Figure 2. Pilot-scale air-stripping column

(1) The turbidity of the water was measured in Nephelometric turbidity units (NTUs) using a Hach model 16800 portable water analysis turbidimeter (Hach Company, Ames, IA). The turbidimeter was calibrated using standards purchased from Hach specifically for this instrument.

(2) The hardness of the water was measured using a Hach Total Hardness Test Kit Model HA-DT (Hach Company, Ames, IA), and read as mg/L $CaCO_3$.

(3) The dissolved solids were measured with a dissolved solids meter which was manufactured by Lamotte Chemical Products Company, Chestertown, MD. The measurements were made in PPMs total dissolved solids.

(4) The chlorine levels were measured using a Wallace and Tiernan (Belleville, NJ) amperometric titrator and reported in mg/L.

(5) The pH of the water was measured using a Corning Science Products (Medfield, MA) Model 130 pH meter.

(6) The color of the water was measured using a Hach Company (Ames, IA) Color Test Kit Model CO-1.

(7) The temperature of the water was measured by drawing off a sample of influent water and immediately inserting a thermometer.

The water samples for these tests were taken in a one-liter flask which was cleaned in chromic acid solution (Manostat Chromerge, New York, NY) and rinsed with distilled water prior to use.

TCE Detection:

The TCE levels in the water were detected using a Hewlett-Packard Model 5840A gas chromatograph (GC) in conjunction with a Hewlett-Packard Model 7675A purge and trap sampler. The purge vessel used was a tube calibrated at 50 mL, the trap used was packed with Tenax GC. The GC was equipped with two 20-ft stainless steel columns packed with 10% SP-1000 supelcoport (Supelco, Inc., Bellefonte, PA) and employed a Dual Flame Ionization Detector.

The water used for making standard solutions and rinsing glassware was deionized water that had been boiled and purged with ultra pure nitrogen gas. The deionized water was supplied by a Milli-RO2575 system (Millipore Corp., Bedford, MA).

All glassware used was cleaned with chromic acid solution and rinsed with nitrogen purged water prior to each use.

The TCE standard curve was prepared by running samples with concentrations of 100, 70, 50, 20, and 10 µg/L (PPB) to insure that the detection of TCE was linear. Samples of approximately 1500 mL were taken from the influent and effluent waters, and from these, 50 mL was used for TCE detection with the purge and trap sampler. For each sample tested, a TCE standard sample was tested as well.

The GC parameters used were: oven temp 120°C, FID temp 225°C, Auxtemp 150°C, Desorb temp 180°C, carrier gas flow 30.0 mL/min., and purge gas flow 20.0 mL/min.

Microbial Identification:

The amount of microbial growth on the lab-scale column was measured weekly. Samples of influent and effluent waters were taken to measure the microbial levels going in and coming out of the column, and samples of the saddles were taken to measure the growth on the column.

The water samples were collected in sterile milk dilution bottles which contained 0.1 mL of 10% sodium thiosulfate to inactivate any residual chlorine that might be present in the water system. The saddles were sampled by randomly selecting five saddles from the top 2.5 cm of the column using sterile forceps and placing them into a test tube containing 5 mL of sterile phosphate buffer and 0.1 mL of 10% sodium thiosulfate.

The test tubes were vortexed for 15 seconds to loosen bacteria from the saddles, then the buffer and the water samples were plated out using the standard plate count method.[11] After the buffer samples were plated out, the excess was decanted off and the saddles were plated out using plate count agar to measure the growth which remained. Since the water in the column was found to be around 16°C, the plates were incubated at 25°C rather than 37°C as specified in standard methods in case there were organisms present which were inhibited by the higher temperature.

The plates were read after 48 hours of incubation at 25°C and distinct colonies were picked on the basis of morphology and color. These colonies were streaked onto plate count agar, then isolated colonies were picked for gram staining, catalase tests, oxidase tests, and for the inoculation of motility medium and two brain heart infusion plates. One of the brain heart infusion plates was incubated aerobically at 25°C; the other was incubated in an anaerobic jar at 25°C.

Identification of the organisms was made using Bergeys Manual of Determinative Bacteriology.[12]

Additional growth tests for presumptive identification of organisms were lactose broth to test for coliforms, mannitol mineral salts[13] agar to test for azotobacter, starch agar, nitrate broth,[14] nitrate reduction test, tryptophane broth to test for indol production, MR-VP broth,[14] Triple sugar iron agar, macConkey agar[15] for detection of coliforms, and API20E diagnostic test strips to identify gram negative enterobacteriaceae.

The 6-ft x 6-in (182.88-cm x 15.24-cm) air-stripping column was measured weekly for the standard plate count in the influent and effluent water. Before the column was shut down and dismantled, a sample of the effluent

water was taken, then the influent water was turned off until the column was drained and then turned on again to flush some of the fouling material from the saddles. A water sample containing the fouling material was collected for chemical and microbial testing.

Iron Fouling Detection:

The method used to detect if iron fouling was present on air stripping columns was a modification of that outlined by Pelosi and McCarthy[16] to detect iron fouling of ion exchange resins, and that outlined in Industrial Inorganic Analysis.[17]

One of the commonly known sources of fouling on the surfaces of water treatment devices is ferric hydroxide.[16,18,19] This compound precipitates out as a gelatinous slime when ferrous iron, which is common in many water supplies, is oxidized.

The test for detection of ferric iron was accomplished by using ammonium thiocyanate to cause a color change in a water sample.

The test used three nessler tubes which were cleaned with chromic acid solution and rinsed with deionized water. To each tube was added 5.0 mL of concentrated HCl and 10.0 mL of a 0.92% NH_4CNS solution. To one tube was added 35.0 mL of effluent water sample and to the remaining two of the tubes was added 35.0 mL of deionized water. One of the tubes containing deionized water served as an iron free control. The second tube was for a comparison with the amount of iron present in the effluent water sample. The comparison was accomplished by adding dropwise, a standard $Fe_2(SO_4)_3$ solution until the color matched that of the effluent sample. The $Fe_2(SO_4)_3$ solution was made by dissolving 0.4474 grams $Fe_2(SO_4)_3$ in 250 mL deionized water which was acidified with a couple of drops of H_2SO_4.

The effluent water from the lab scale column was tested for iron levels as well as the two samples of effluent water from the pilot scale column. One of the two samples from the pilot scale column was taken from the effluent water after the influent water was turned off and then turned on again to flush some of the slime material from the saddles.

Disinfectants:

(1) Disinfectant, Food Service

Samples of the ceramic saddles, which were received from ESE as packing material for the columns, were plated out with plate count agar and found to have a heavy microbial load on them. The saddles used in the laboratory scale column were used as they were received, however the saddles used in the pilot scale column were sterilized before use.

The saddles were sterilized by soaking them in a concentrated solution of Disinfectant, Food Service Experimental Formulation 3-A (meets military standard MIL-D-11309F) for one week. The disinfectant has potassium dichloroisocyanurate as the active ingredient and was prepared by dissolving one 4.77-oz. (135.13-gm) packet into 20 liters of water, giving a chlorine concentration of approximately 770 ppm.

After one week in the disinfectant, five of the saddles were aseptically removed from the disinfectant and placed into 5.0 mL of sterile phosphate buffer containing 0.05% sodium thiosulfate to neutralize the chlorine. The buffer solution and the saddles were plated out on plate count agar to test for microbial growth.

(2) Hydrogen Peroxide

After the laboratory scale column was operated for approximately 5 1/2 months, an investigation was conducted into the use of Hydrogen Peroxide (H_2O_2) for disinfection purposes.

The method used to sample was as follows: Five saddles were removed aseptically from the column and placed in a sterile test tube containing 5.0 mL of phosphate buffer, 1.0 mL of 10% catalase, and 0.1 mL of 10% sodium thiosulfate. Samples of 10.0 mL each were taken from the liquid at the top of the column and from the effluent hose. These samples were taken in sterile test tubes containing 1.0 mL of 10% catalase and 0.1 mL of 10% sodium thiosulfate. The catalase used was filter sterilized using a 115-ml (0.45-μm) millipore filter.

Each of the samples were plated out using plate count agar and incubated at 25°C. The presence of residual peroxide in the samples was tested for by using potassium iodide starch paper.

Before the column was treated with peroxide, microbial test samples were taken, then the air and effluent water hoses were clamped off.

The column was first flooded with 3% H_2O_2 and microbial test samples taken at one-hour intervals for five hours. The column was drained after two hours and replaced with fresh solution of 3% H_2O_2 for the remaining three hours.

The day after the column was treated with 3% H_2O_2, the column was operated and samples were taken from the saddles and from the effluent water.

The column was operated for 3 more days and then treated with a 15% H_2O_2 solution for 24 hours. Samples were taken from the liquid at the top of the column and saddles as before at hours 1, 2, 3, 4, 8, and 24.

RESULTS

Slime Buildup on Columns:

Both the laboratory and pilot scale air stripping colums experienced slime buildup after a few weeks of operation. The type of fouling for each

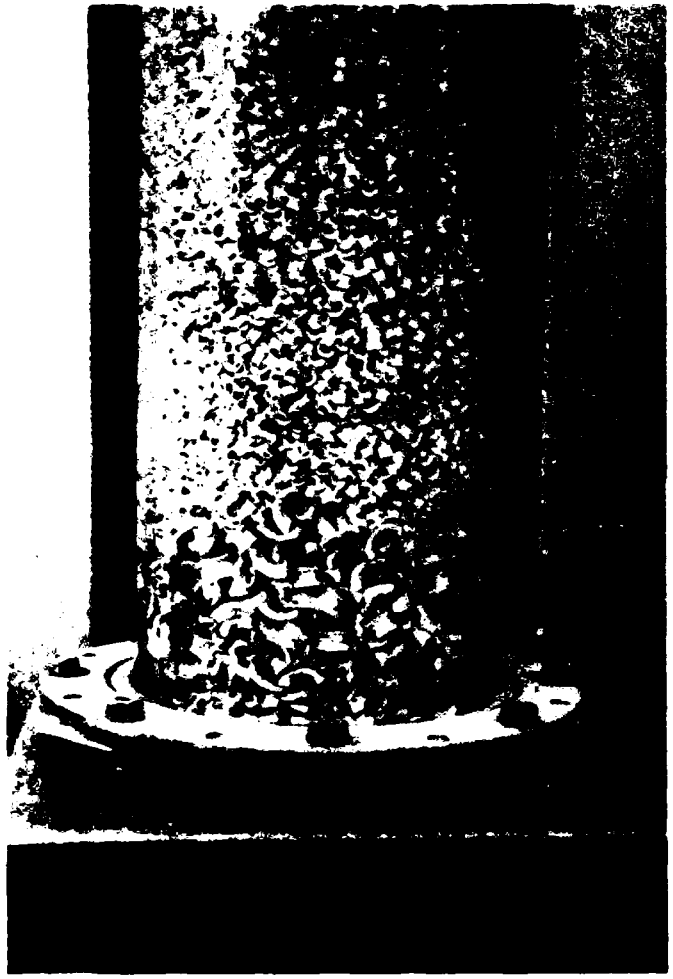

Figure 3. Saddles in pilot-scale air-stripping column before use.

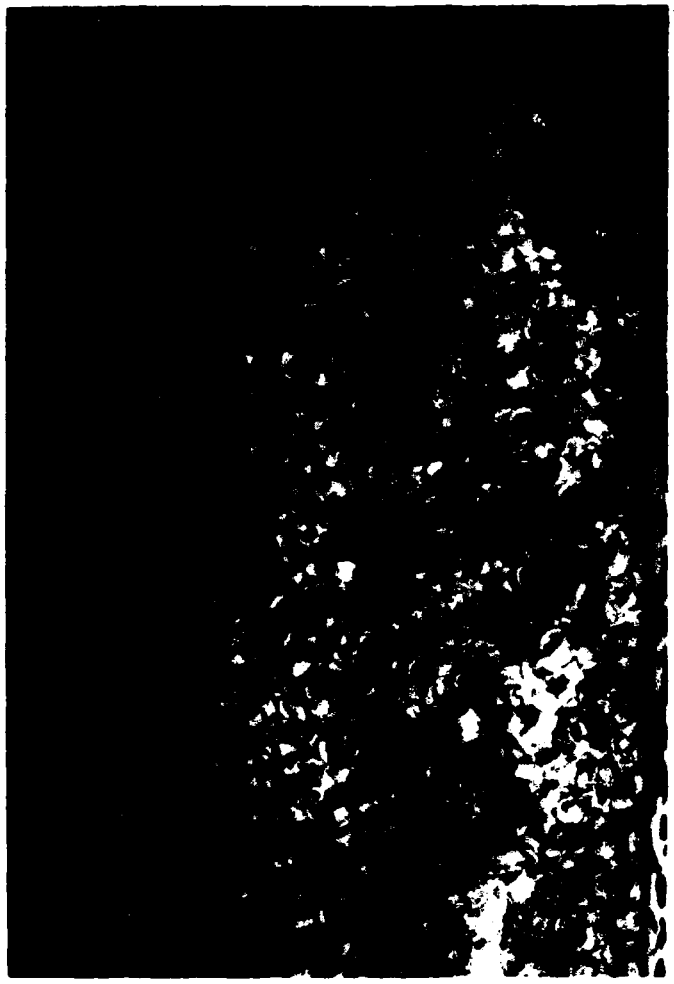

Figure 4. Saddles in pilot-scale air-stripping column showing gelatinous slime buildup after 3 wks operation.

TABLE 1. Weekly Characterization Data for Laboratory-Scale Air-Stripping Column Influent Water.

Week	1	3	4	5	6	7
Date	7/12/84	7/23/84	7/31/84	8/ 7/84	8/14/84	8/24/84
Temp (^{o}C)	15.6	17.6	15.9	17.8	16.6	16.4
Hardness (mg/L)	150.0	144.0	132.0	130.0	142.0	136.0
Turbidity (NTU)	0.3	1.2	1.4	2.1	1.4	0.50
pH	6.6	6.6	6.7	6.7	6.8	6.7
Color (CPU)	0.0	0.0	0.0	0.0	0.0	0.0
Chlorine (PPM)	0.0	0.0	0.0	0.0	0.0	0.0
Total Dissolved Solids (PPM)	- *	-	-	-	-	-

Table 1. Cont'd.

Week	9	10	16	17	18	19
Date	9/ 5/84	9/13/84	10/26/84	11/ 2/84	11/ 9/84	11/21/84
Temp (^{o}C)	15.8	15.9	12.0	16.1	15.9	15.5
Hardness (mg/L)	142.0	129.0	130.0	135.0	131.0	133.0
Turbidity (NTU)	0.40	0.21	0.22	0.41	0.69	0.35
pH	6.8	6.5	6.6	6.6	6.8	6.8
Color (CPU)	0.0	0.0	0.0	0.0	0.0	0.0
Chlorine (PPM)	0.0	0.0	0.0	0.0	0.1	0.12
Total Dissolved Solids (PPM)	-	300.0	310.0	300.0	300.0	310.0

Table 1. Cont'd.

Week	20	21	22	23	24
Date	11/30/84	12/ 6/84	12/14/84	12/20/84	12/27/84
Temp (^{o}C)	15.0	14.5	15.0	14.7	15.0
Hardness (mg/L)	125.0	129.0	122.0	136.0	129.0
Turbidity (NTU)	0.34	0.34	0.52	0.31	0.70
pH	6.8	6.8	6.9	6.9	6.9
Color (CPU)	0.0	0.0	0.0	0.0	0.0
Chlorine (PPM)	0.11	0.11	0.15	0.09	0.05
Total Dissolved Solids (PPM)	330.0	310.0	310.0	300.0	290.0

* - No data

will be discussed later. Figures 3 and 4 show the pilot scale air stripping column before and after use. After three weeks of operation, the saddles became coated with a brown gelatinous layer on areas where the water flowed.

Water Characterization:

The results of the water characterization tests for the air stripping columns influent waters are listed in Tables 1 and 2.

The characterization values remained fairly constant over the time period tested. Both columns received water which was from the same source and had no significant differences other than temperature. The pilot scale column was operated outside where the surrounding temperature was lower than that around the lab scale column.

TABLE 2. Weekly Characterization Data for 6-ft x 6-in Air-Stripping Columns Influent Water

Week Date	1 11/28/84	2 12/5/84	3 12/12/84	4 12/19/84
Temp (^{o}C)	13.0	13.1	12.6	12.8
Hardness mg/L	123.0	133.0	130.0	143.0
Turbidity (NTU)	0.38	0.21	0.51	0.50
pH	6.8	6.9	6.9	6.8
Color	0.0	0.0	0.0	0.0
Dissolved Solids (PPM)	300.0	300.0	300.0	290.0
Chlorine (PPM)	0.15	0.10	0.06	0.16

TCE Detection:

When operation of the lab scale air-stripping column began, the TCE levels of the influent and effluent waters were measured with a gas chromatograph equipped with a purge and trap sampler and a dual flame ionization detector (Table 3).

Two weeks after the column operation began, the gas chromatograph began to malfunction. While awaiting repair of the GC, weekly water samples were taken and refrigerated for later analysis.

As the microbial levels on the column increased, decreased column efficiency became a concern. Since there were no means to monitor the TCE levels in the effluent water, the spiking of the column with TCE was terminated to prevent discharge of the chemical into the sewer system.

TABLE 3. TCE Concentration from Laboratory Scale Air-Stripping Column Influent and Effluent Water Samples

Week No	Sample	TCE Area	TCE Conc.	mL/min H_2O
1	influent	202,200	300 PPB	1167
	effluent	0	<0.0014 PPB *	1167
2	influent	159,200	236 PPB	1800
	effluent	9,184	14 PPB	1800
Standards				
		68,980	100 PPB	
		47,310	70 PPB	
		32,880	50 PPB	
		17,460	30 PPB	
		13,530	20 PPB	
		7,320	10 PPB	

* Detection limit based on 10 PPB standard.

Biogrowth on Pilot-Scale Air-Stripping Column:

The pilot-scale 6' x 6" air-stripping column had a heavy layer of slime accumulating on the saddles after four weeks of operation. The results of the microbial testing of the effluent water, shown in Table 4, indicate very little biogrowth on the column.

TABLE 4. Average Standard Plate Count/mL of Water Samples from Pilot-Scale 6-ft x 6-in Air-Stripping Column

	Weeks			
Sample	1	2	3	4
influent water	1.0 cfu	6.5 cfu	1.0 cfu	0.0 cfu
effluent water	8.0 cfu	7.5 cfu	7.0 cfu	0.5 cfu

At the end of the study, a sample of the effluent was taken just before the influent water was turned off. The influent was turned on again to flush some of the slime material from the column, and another effluent water sample was taken. The results of the microbial tests of these samples shown in Table 5 indicate that the slime material was not caused by microbial growth.

Table 5. Standard Plate Count/mL of Effluent Water Containing Slime Material from Pilot-Scale 6-ft x 6-in Air-Stripping Column

Sample	Standard Plate Count/mL
effluent water before flushing	130
effluent water after flushing	12

Biogrowth on Laboratory-Scale Air-Stripping Column:

The amount of biogrowth on the lab scale column increased significantly from the time that the operation began. The results in Table 6 show the average standard plate count of organisms present on the saddles and in the influent and effluent waters during the weekly samplings. The values obtained for the saddles are the number of bacteria released from the saddles into 5 mL of buffer solution after mixing for 15 seconds. These values should only be compared with other sample values for saddles and not with the effluent water samples. Since the sampling procedure was constant, the values should indicate the relative change of growth over time.

The predominant types of bacteria isolated from the column were Pseudomonas, Bacillus, and Azotobacter. Other types of organisms isolated in lesser amounts were; Actinomyces, Flavobacterium, Staphylococcus, Beijerinckia, Zoogloea, and Nitromonas.

The criteria used in making the identification of the organisms were as follows:

Pseudomonas: Gram negative rods, motile, catalase positive, oxidase positive, strict aerobes, no starch hydrolysis, no nitrate reduction, no growth on mannitol mineral salts medium.

Bacillus: Gram positive rods, central spore, motile, catalase positive.

Azotobacter: Large ovoid cells, produce copious amounts of slime, Gram variable, growth in mannitol mineral salts medium, hydrolyses starch, catalase positive, motile, grew in plate count broth containing 5% ETOH, one strain produced fluorescent green pigment under UV light.

Actinomyces: Branched filament, one colony white crusty, one yellow, thick and moist.

TABLE 6. Average Standard Plate Count/mL of Samples from Laboratory-Scale Air-Stripping Column.

WEEK	INFLUENT	EFFLUENT	SADDLES
1	109.0	420.0	73.5
2	18.0	24.5	2190.0
3	194.0	7600.0	1450.0
4	2.0	995.0	7800.0
5	155.0	27600.0	10900.0
6	2.5	9600.0	20200.0
7	1.0	1640.0	18000.0
8	2.5	98.5	7250.0
9	0.5	1165.0	13850.0
10	0.5	7000.0	56500.0
11	19.0	1935.0	18400.0
12	0.0	530.0	2500.0
13	9.5	660.0	38500.0
14	25.5	5750.0	94500.0
15	0.0	2070.0	515000.0
16	2.0	980.0	103500.0
17	29.0	510.0	51000.0
18	45.5	2935.0	53000.0
19	52.0	775.0	28050.0
20	15.5	690.0	87500.0
21	0.5	385.0	103500.0
22	5.5	2150.0	122500.0
23	6.0	3000.0	239000.0
24	53.0	760.0	77000.0
25 *	4.0	40000.0	13600.0
27 **	--	--	19800.0

* One week after treatment with 3% H_2O_2

** Three weeks after treatment with 15% H_2O_2

Flavobacterium: Slender Gram negative rods, yellow pigment, difficult to maintain in culture, growth inhibited at 37°C.

Staphylococcus: Gram positive cocci, growing in clumps.

Beijerinckia: Gram negative rods, strict aerobes, motile, catalase positive, produced a tough elastic slime, grew on mannitol mineral salts medium, hydrolysed starch, grew on plate count broth containing 5% ethanol.

Zoogloea: Gram negative rods, motile , strict aerobe, oxidase positive, catalase positive, citrate not utilized, indole not produced, grew on 3% NaCl agar but not 6% NaCl agar, flocs formed when grown in nutrient broth, older cells distended (possibly due to poly B-hydroxybutyric acid).

Nitromonas: Short gram-negative rods, strict aerobes, grew in short chains, cells yellow and embedded in a slime matrix, grew on basal salts medium with *ammonia (chemolithotroph)*.

Iron Fouling Detection:

The slime buildup on the pilot-scale 6-ft x 6-in air stripping column was determined to be caused by iron compounds rather than by microbial growth, based on the test for iron and the standard plate count tests.

When 35.0 mL of effluent water containing the slime material was flushed from the column, mixed with 5.0 mL of HCl and 10.0 mL of 0.92% NH_4CNS solution, a red color developed. The comparator tube required 15 drops of standard $Fe_2(SO_4)_3$ solution to produce the same color reaction. The volume of $Fe_2(SO_4)_3$ used was approximately 0.75 mL which gives a value of 10.714 ppm in the 35.0-mL sample.

The effluent water sample taken before flushing the slime from the column gave no detectable iron using this method of analysis.

The analysis of the effluent water from the lab scale column indicated that iron fouling was present although at a lesser degree. The effluent

water with material flushed from the column caused a red color reaction but not as dark as the 6-ft x 6-in column effluent. The comparator tube required 5 drops of standard $Fe_2(SO_4)_3$ solution to equal the color change of the effluent water. This volume of $Fe_2(SO_4)_3$ indicated a concentration of 3.571 ppm in the effluent water, a value of one-third that of the 6-ft x 6-in column.

Disinfection of Saddles Prior to use in the 6-ft x 6-in Pilot-Scale Column:

The saddles which were plated out on plate count agar before the treatment with the potassium dichloroisocyanurate based disinfectant had a heavy overgrowth of bacteria on the plate (shown in Fig. 5). After the saddles were soaked in the disinfectant for a week there was no sign of microbial activity on them (Fig. 6). The saddles were bleached out by the disinfectant and showed no trace of organic or other debris attached to the surface.

Results of Hydrogen Peroxide on Biogrowth in Laboratory-Scale Column:

Although the use of hydrogen peroxide as a disinfectant is well documented,[20-24] the treatment of the lab-scale air-stripping column with hydrogen peroxide resulted in a slight decrease in the amount of bacteria present, but it did not disinfect the column.

The treatment with 3% H_2O_2 killed off all species of bacteria except for Psuedomonas and Bacillus. These were two of the most predominant organisms found on the column during operation so the total plate count was not greatly reduced by the peroxide. The results of the microbial sample taken during treatment with 3% H_2O_2 are listed in Table 7.

The treatment with 15% H_2O_2 failed to remove the Psuedomonas from the column but it did seem to remove the vegetative Bacillus cells. The

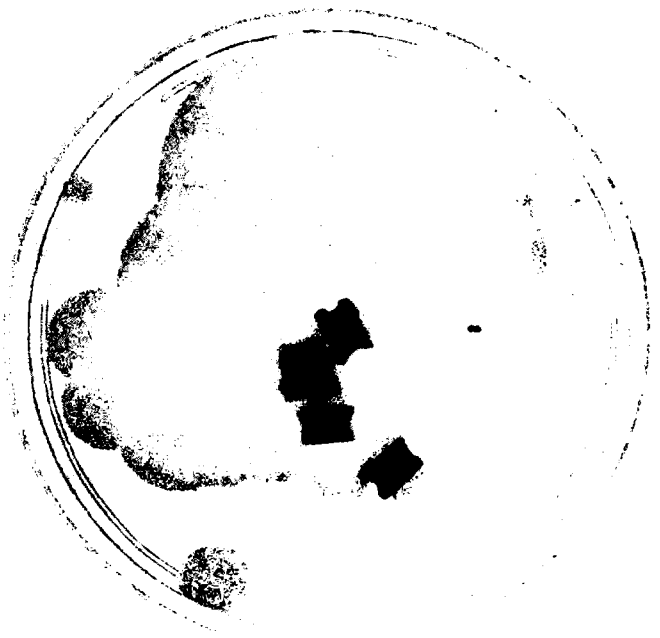

Figure 5. Saddles on SPC agar before treatment with Disinfectant, Food Service Experimental Formulation 3-A

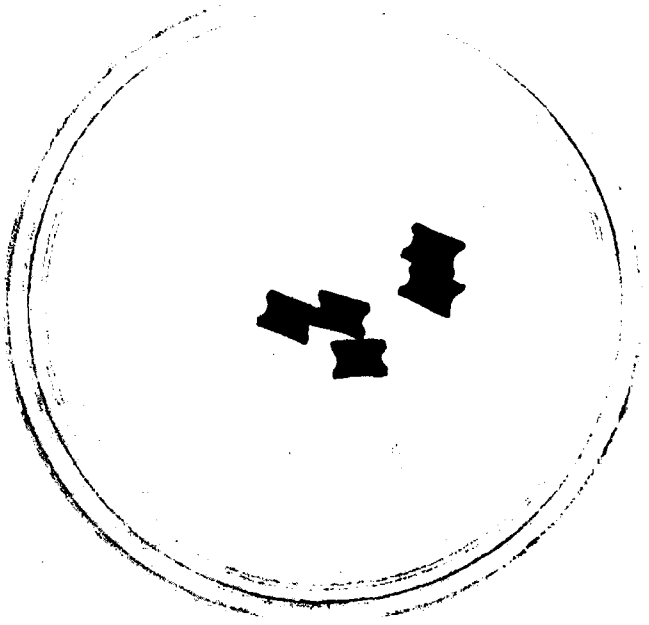

Figure 6. Saddles on SPC agar after treatment with Disinfectant, Food Service Experimental Formulation 3-A

results of the microbial test samples taken during the treatment with 15% H_2O_2 are listed in Table 8.

The column was operated for 3 weeks after treatment with 15% H_2O_2 and sampled for measurement of bacterial regrowth. The sample revealed a regrowth of Bacillus on the saddles at a concentration of 1.98×10^4/mL. This indicates that some of the Bacillus spores may have survived the treatment with 15% H_2O_2.

TABLE 7. Average Standard Plate Count/mL (in hours) During Treatment with 3% H_2O_2

Sample	1	2	3	4	5
Top of Column	118.5	12.5	0	2.5	16
Liquid Effluent	0	0	23	55.5	47.5
Saddles	134	121	TNTC *	TNTC	TNTC

*- Too numerous to count

TABLE 8. Average Standard Plate Count/mL (in hours) During Treatment with 15% H_2O_2

Sample	1	2	3	4	8	24
Top Liquid	1.5	135.5	135	159.5	107.5	108
Effluent	0	0	0	0	0	0
Saddles	138	154	148.5	141.5	163.5	144

DISCUSSION

In the operation of the pilot-scale 6' x 6" air-stripping column, iron fouling was found to be a greater problem than was biogrowth.

After four weeks of continuous operation, the packing material in the column was coated with iron compounds, presumably ferric hydroxide; whereas

the effluent water samples containing that fouling material showed little microbial activity.

The well-water which was used for operation of the column was known to have high iron content so the fouling of the column with iron compounds was not unexpected.

The low incidence of microbial fouling on this column may have been partly due to the water temperature (13^{o}C) inhibiting growth of many organisms, and to the lack of inoculum due to the sterilization of the saddles before use. Also, during the operation of the column there was air exiting through the top of the column, creating a positive pressure effect which would decrease the chance of contamination from above. Any bacterial contamination would have to enter through the water or the air.

The recommended method for cleaning the iron fouling material from the column is by using dilute (10%) hydrochloric acid. This method has been successfully employed for cleaning iron fouling material from ion exchange resins [16,18,19] and membrane processing and has also been shown to reduce bacterial numbers.[16]

Other methods employed for this purpose are the use of sequestrants and inhibited acids.[18,25]

During operation of the laboratory scale column, biogrowth was much more prevalent, and iron fouling appeared to be less evident. There are a number of factors that could account for the increased biogrowth on this column.

The first factor was the presence of biogrowth on the saddles before use. When the packing material was received from ESE, the Flora remaining from previous usage served as an incculum for this column.

The second important factor was the schedule of operation. This possibly increased the bacterial growth, because when the column was not in operation,

the temperature inside the column increased and there was no mechanical shearing force from the water to dislodge the microbial flora.

There have been reports of chlorine resistant organisms,[26,27] specifically Bacillus,[28] and Pseudomonas.[27,29,30] One proposed method of bacterial resistance to chlorine is by means of an extracellular mucopolysaccharide slime layer[28,30,31] which in this case could also cause fouling of the columns. It was reported[30] that slug chlorination of cooling towers removed some bacteria but regrowth occured in a few hours.

When the saddles to be used in pilot-scale 6-ft x 6-in air stripping column were soaked in potassium dichloroisocyanurate, there were no signs of regrowth of Pseudomonas or Bacillus after a month of continuous operation.

Chlorine demand can be exerted by inorganic ferrous, manganous nitrate, and sulfate ions[28] as well as organic debris. For this reason, the air-stripping column should be cleaned of the iron fouling material prior to chlorination.

The chlorination of water contaminated with organic or humic matter has been implicated in the production of haloforms [32,33] and organic halides.[32-34]

Three other compounds cited for use as disinfectants in water systems were ozone,[25,34,35] formaldehyde,[16] and quanternary ammonium compounds.[36]

During the course of the study with the laboratory scale air-stripping column, the TCE spike that was used to simulate contaminated well-water was discontinued because of a malfunction in the gas chromatograph which was used for TCE detection and measurement.

It is difficult to speculate whether the TCE had any significant effect on the biogrowth, since it was not spiked into the entire length of the

study. Because biogrowth occurred on the column during the month that it was being spiked, it seems doubtful that TCE had any deleterious effect on growth, although one study[37] has shown Pseudomonas putida to be inhibited by TCE.

Although some recent studies[38] have shown that TCE can be degraded aerobically when exposed to a mixture of natural gas in air, most sources[39] consider TCE to be nonbiodegradable under aerobic conditions. Since TCE is not considered biodegradable under these conditions, there is no reason to believe that its presence would have enhanced bacterial growth on the column.

CONCLUSIONS and RECOMMENDATIONS

The results of this study indicate that iron fouling of air-stripping columns is a more common problem than biofouling.

The procedure recommended for correction or prevention of iron fouling is to flush the columns with dilute hydrochloric acid.

The procedure recommended for the prevention of biogrowth is (1) to sterilize the packing material before use, (2) operate the column continuously to keep positive air pressure at the top of the column, and (3) keep the internal temperature of the column as low as possible.

The column should be monitored for biogrowth and if a problem should occur, then super chlorination should be implemented immediately subsequent to iron removal.

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